INTRODUCTION
Pregnancy, also known as gravidity or gestation, is the time during which one or more offspring develops inside a woman. During pregnancy, the woman undergoes deep anatomical, physiological, biochemical and endocrine changes that affect multiple organs and systems. These changes are essential to help the woman to adapt to the pregnant state and to aid fetal growth and survival. However, such anatomical and physiological alterations may cause confusion at some stage in clinical examination of a pregnant woman. Pregnancy typically divided into three trimesters is characterized by physiological changes which are reversible while Pathologic variations of these changes are not, thus persisting even after pregnancy. Successful outcome of pregnancy requires frequent monitoring of biochemical and hematological parameters to avoid complications throughout the trimesters of pregnancy.[1]

Similarly, changes in blood biochemistry during pregnancy may create difficulties in interpretation of results. Conversely, clinicians also need to recognize pathological deviations in these normal anatomical and physiological changes during pregnancy to institute appropriate action to improve maternal and fetal outcome.[1]
Urea is the principal nitrogenous waste product of metabolism and is generated from protein breakdown. It is eliminated from the body almost exclusively by the kidneys in urine, and measurement of its concentration, first in urine and later in blood, has had clinical application in the assessment of kidney (renal) function. Urea owns special historical significance compared with most other analytes currently measured in the clinical laboratory or at the point of care.\textsuperscript{[23]}

Any pathology associated with tissue breakdown is for the same reason associated with increased urea production. A small amount (<10 %) of urea is eliminated via sweat and the gut, but most of the urea produced in the liver is transported in blood to the kidneys where it is eliminated from the body in urine. The process of renal elimination, starts with filtration of blood at the glomeruli of the roughly 1 million nephrons enclosed within each kidney. During glomerular filtration, urea passes from blood to the glomerular filtrate, the fluid that is the precursor of urine. The concentration of urea in the filtrate as it is formed is similar to that in plasma so the amount of urea entering the proximal tube of the nephron from the glomerulus is determined by the glomerular filtration rate (GFR).\textsuperscript{[3]}

Creatinine is a chemical waste product that is produced by the muscle metabolism and to a smaller extent by eating meat. Healthy kidneys filter creatinine and other waste products from your blood. The filtered waste products leave the body in urine. An increased level of creatinine may accumulate in the blood if the kidney is malfunctioning.\textsuperscript{[4]}

**MATERIALS AND METHOD**

**MATERIALS**

Chemicals and reagents used include the following: 14% Sodium Carbonate, 50% Acetic acid, conc. \(\text{H}_2\text{SO}_4\), Phosphoric Acid, Diacetyl monoxide thiosemicarbazide, phenyl mercuric acid, 10% Sodium tungstate, Lithium carbonate, N/12 \(\text{H}_2\text{SO}_4\), Phosphotungstic acid, \(\text{FeCl}_3\cdot6\text{H}_2\text{O}\), distilled water, Analar grade picric acid, 0.75 N Sodium hydroxide and other widespread laboratory reagents. The instruments used include spectrophotometer, refrigerator, water bath, centrifuge and other common laboratory apparatus.

**STUDY POPULATION**

A total of 100 subjects were used for this study; 75 were pregnant women attending ante-natal clinic in Rumuolumeni Health Center, Rumuolumeni, Ohio/Akpor, Rivers State, Nigeria, while the other 25 women were randomly selected non-pregnant women within the clinic environment, which served as control. The pregnant women were divided into three groups of 25 each, based on the trimester of their pregnancies. Relevant information about pregnancy and health status of the subjects was obtained, after they gave their informed consent.

**COLLECTION OF SAMPLES**

A 5 ml blood was collected by venous puncture using a 5 ml sterile syringe and needle. The blood was dispersed into clean dry tubes, allowed to clot for about 15 minutes at room temperature, then centrifuged at 3000rpm for 5 minutes and the serum was harvested into clean dry screw-capped bottles.

**BIOCHEMICAL ASSAY METHODS**

**Estimation of Serum Urea**

Venous blood samples were collected from antecubital vein after an overnight fasting from all participant’s with aseptic precautions. The method of estimation of serum urea using thiosemicarbazide is the standard Diacetyl monoxime method. There are 2 stages in this technique, which includes the following:

**Stage I**: test tubes were labeled test, standard and blank. Then 10 ml of distilled water was added to the tubes labeled test and standard, followed by 0.1ml serum placed in the tube labeled test while 0.1 ml urea standard was added to the tube labeled standard and each was thoroughly mixed.

**Stage II**: 1 ml of diluted serum was transferred into another tube labeled test, followed by 1 ml of distilled water, 2ml urea acid reagent and 2ml urea colour reagent.

To another tube labeled standard, 1ml diluted standard was placed, followed by 1 ml distilled water, 2ml each of urea acid and urea colour reagents. To the tube labeled blank, 2ml distilled water was added, followed by 2ml each of urea acid and urea colour reagents. All the tubes contents were thoroughly mixed, then placed in a boiling water bath at 100\(^\circ\)C for 20 minutes; removed, allowed to cool and absorbance was read at 520 nm against reagent blank. The concentration of urea was calculated by the expression:

\[
\text{Urea (mmol/l)} = \frac{\text{Absorbance of test tube}}{\text{Absorbance of standard}} \times \text{Concentration of standard (10 mmol/l)}
\]

Where A represent absorbance.

**Estimation of Serum Creatinine by Jaffe’s Method**

This technique also has 2 stages:

**Stage I**: test tubes were labeled test, standard and blank. To the test, 3ml distilled water, 1ml of serum, 1ml 10 % sodium tungstate and 1ml \(\text{H}_2\text{SO}_4\) were added. To the blank was added 4 ml distilled water, 1ml 10 % sodium tungstate and 1 ml 2/3N \(\text{H}_2\text{SO}_4\). Each tube content was mixed and centrifuged for 5 minutes at 3000rpm.

**Stage II**: 3ml of each supernatant from stage I was added to fresh test tubes labeled test and blank while 3ml of creatinine standard was added to tube labeled standard. To the test, standard and blank was added 1 ml each of 0.75 N NaOH and Picric acid. These were mixed thoroughly and allowed to stand for 15 minutes at room temperature, then optical densities of test and standard were measured using spectrophotometer at 520nm, after
The concentration of creatinine was calculated by the expression:

\[ \text{Creatinine (μmol/l)} = \frac{A_{\text{std}}}{A_{\text{obs}}} \times \text{Concentration of standard (530 μmol/l)} \]

RESULTS

The results of the experiments were presented in tables. Table 1 shows the mean levels of serum creatinine in pregnant women attending ante-natal clinic at Rumuolumeni Health Center, Rumuolumeni, Obio/Akpor, Rivers State, Nigeria, at different trimesters of pregnancy and non-pregnant women as control. These results showed that there was a progressive decrease in the levels of serum creatinine in the first trimester. The second semester had a significant increase in the second trimester, but the third trimester showed a significant \( (p<0.05) \) decrease in the levels of serum creatinine when compared with the control.

Table 1: Table Showing Levels Serum Creatinine in Pregnant Women at Rumuolumeni Health Center, Rumuolumeni, Obio/Akpor, Rivers State, Nigeria.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control</th>
<th>1st trimester</th>
<th>2nd trimester</th>
<th>3rd trimester</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum Creatinine (μmol/l)</td>
<td>77.50</td>
<td>70.75 ±23.20</td>
<td>78.50 ±18.30</td>
<td>73.45 ±14.98</td>
</tr>
<tr>
<td>Serum Urea (mmol/l)</td>
<td>4.11 ±0.71</td>
<td>3.60 ±0.72</td>
<td>3.49 ±0.80</td>
<td>3.29 ±0.78</td>
</tr>
</tbody>
</table>

Values are means±SD, where SD=standard deviation, \( n=100 \).

Table 2 shows the age distribution of pregnant women across three trimesters, attending ante-natal clinic at Rumuolumeni Health Center, Rumuolumeni, Obio/Akpor, Rivers State, Nigeria, and the control. This shows that the highest percentage of pregnant women (33%) were from age group, 26 – 30, while the lowest percentage of pregnant women (6%) were from age group, 36 – 40.

Table 2: Age distribution of pregnant women across three trimesters, attending ante-natal clinic at clinic at Rumuolumeni Health Center, Rumuolumeni, Obio/Akpor.

<table>
<thead>
<tr>
<th>Age Group</th>
<th>1st trimester</th>
<th>2nd trimester</th>
<th>3rd trimester</th>
<th>Control</th>
<th>% Age</th>
</tr>
</thead>
<tbody>
<tr>
<td>16 - 20</td>
<td>7</td>
<td>4</td>
<td>8</td>
<td>8</td>
<td>27</td>
</tr>
<tr>
<td>21 - 25</td>
<td>3</td>
<td>3</td>
<td>5</td>
<td>2</td>
<td>13</td>
</tr>
<tr>
<td>26 - 30</td>
<td>9</td>
<td>11</td>
<td>7</td>
<td>6</td>
<td>33</td>
</tr>
<tr>
<td>31 - 35</td>
<td>4</td>
<td>6</td>
<td>3</td>
<td>8</td>
<td>21</td>
</tr>
<tr>
<td>36 - 40</td>
<td>2</td>
<td>1</td>
<td>2</td>
<td>1</td>
<td>6</td>
</tr>
</tbody>
</table>

DISCUSSION

The results of this study concur with the known physiological changes of pregnancy; namely a fall in serum creatinine due to gestational hyper filtration resulting in a 50% increase in creatinine clearance by the second trimester \(^{[5,6,7]} \) followed by a decrease in creatinine clearance during the third trimester \(^{[8]} \) leading to an increase in serum creatinine concentration toward term.

Acute kidney injury occurs most commonly during pregnancy in the third trimester, predominantly due to the development of hypertensive disorders and puerperal pathologies including sepsis and hemorrhage. Diagnostic criteria for acute kidney injury do not exist in pregnancy, and up to 40% of pregnancy-associated acute kidney injury may be missed by clinicians in the Obio/Akpor.

The physiological condition of pregnancy carries about a lot of changes which have an effect on the metabolism of a range of biochemical parameters. These changes are mostly thought to offer conducive environment for the growing fetus but can affect the health of the woman and could also lead to problems with metabolism and excretion of biochemical markers of renal impairment. Furthermore, during pregnancy cardiac output and renal blood flow are increased together with physiological increase in GFR resulting to an increased clearance of creatinine. \(^{[9]} \) To this end, a slight rise in creatinine level during pregnancy may indicate progression of renal disease and thus serum creatinine has greater prognostic ability for the determination of the adverse outcomes of kidney disease. \(^{[10,11,12]} \)

In this study, creatinine concentration was significantly reduced among the pregnant women compared to the non-pregnant controls. This is in consonant with one study which reported significantly lower creatinine levels in the cases as against the control. \(^{[13]} \) Some report indicated about 50% increase in glomerular filtration rate during pregnancy \(^{[14]} \), and this could lead to increase in creatinine excretion. Creatinine is freely filtered and its level falls in normal pregnancy due partly to a pregnancy-induced increase in GFR on one hand and on the other hand due to hemodilution from plasma expansion culminating in the decrease in serum

<table>
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<tr>
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<th>Values</th>
<th>Notes</th>
</tr>
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<tbody>
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<td>Serum Urea (mmol/l)</td>
<td>4.11 ±0.71</td>
<td>Values are means±SD, where SD=standard deviation, ( n=100 ). Values with asterisk(*) are significantly different from the control at ( p&lt;0.05 ).</td>
</tr>
</tbody>
</table>
creatinine concentration.\textsuperscript{[15]} Consequently, the reduction in serum creatinine is ancillary to plasma volume expansion, renal vasodilation, hyperfiltration, and increased glomerular basement membrane permeability.\textsuperscript{[16]} The gradual decrease in the concentration of creatinine in plasma from the first to the third trimesters of pregnancy is likely to be as result of increase in GFR associated with pregnancy but not a reduction in its plasma concentration. The increase in GFR may be due partly to upsurge in the concentration of aldosterone which increases the blood volume, in some instances, up to 50% and increase renal blood flow\textsuperscript{[17]} resulting in an increase in the rate at which creatinine is cleared from plasma.

Urea is the main waste product of protein breakdown. It is synthesized in the liver from ammonia which is toxic to the body, but formed as a result of deamination of amino acids.\textsuperscript{[18]} The decrease in serum urea of pregnant women in all trimesters even though not significant might be due to hydration, a rise in glomerular filtration rate (GFR), increased anabolic rate and demand of the developing foetus on the protein of pregnant mothers. A rise in the GFR was thought to account for the increased excretion of urea. As GFR increases without substantial alteration in urea production, due to limited intake of protein, concentration of this molecule decreases in plasma. The alteration in protein metabolism in late pregnancy suggests that amino acids are conserved for tissue synthesis. The sum total of plasma amino acids decline in pregnancy is between 15 -25\%, reflecting enhanced placental uptake and increased anabolic rate. It is a well known fact that the level of urea in urine acutely decreases when dietary protein is restricted, which is an indication of reduced plasma urea.\textsuperscript{[19]} It appears therefore that as GFR increases in normal pregnancy, in addition to increased anabolic rate, serum concentration of urea decreases.

CONCLUSION

Normal pregnancy is associated with progressive decrease in urea and creatinine levels from the first trimester to the third trimester.

The absence of reliable data on reference intervals for creatinine and Urea among pregnant women in Ohio/Akpor calls for the establishment of these reference ranges using larger sample size and should cover all the regions of the State. This is because the physiological and anatomical changes that come with pregnancy especially those related to the kidney means that the laboratory reference intervals of non-pregnant women are not suitable for pregnant women.

REFERENCES